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VARIATION IN *SCHIZOCOSA* (ARANEAE: LYCOSIDAE), *METAPHIDIPPUS* AND *PHIDIPPUS* (ARANEAE: SALTICIDAE)[1]

William W. M. Steiner and **Matthew H. Greenstone:** Biological Control of Insects Research Laboratory, USDA, Agricultural Research Service, P.O.B. 7629, Columbia, Missouri 65205 USA

Gail E. Stratton: Department of Biology, Albion College, Albion, Michigan 49224 USA

ABSTRACT. Allozyme variation was examined in five species of solitary spiders collected in Illinois and Missouri including *Schizocosa ocreata* (Hentz), *S. stridulans* Stratton, *S. rovneri* Uetz & Dondale, *Phidippus clarus* Keyserling, and *Metaphidippus galathea* (Walckenaer). The average number of alleles per locus was small, and the average heterozygosity ranged from 2.3 to 12.5. The percent of polymorphic loci ranged from 16.6 to 66.7%. For one species, *P. clarus*, a Missouri population is compared to a population from South Carolina (Terranova & Roach 1987). Overall, the genetic variability estimates are lower than those for other North American spiders. However, the observed genetic variation is approximately four times higher than that observed in communal spiders.

Genetic variation is generally considered necessary, if not crucial, to a species' ability to adapt to changing environmental conditions. Recently, Terranova & Roach (1987) reported electrophoretic variation at 12 isozyme loci in 7 species of the solitary spider genus *Phidippus* collected from South Carolina. They found high estimates of variation with 41.6% of loci polymorphic and a mean heterozygosity of 11.7%.

In contrast, Lubin & Crozier (1985) investigated 21 enzyme systems and found only one polymorphic isozyme locus in the social spider *Achaearana wau* Levi of New Guinea. The variation occurred in only 6 of 30 naturally-occurring colonies. Thus 24 of the colonies displayed no polymorphism in the form of variable enzyme systems in the 21 enzymes which were examined.

Lubin and Crozier hypothesized that social spiders will have lower variation as a result of close inbreeding within the eolony. Their work seems to be supported by subsequent studies of social spiders and what is known about inbreeding and genetic variation in eusocial Hymenoptera (Graur 1985; Berkelhamer 1983; Reeve et al. 1983; Owen 1983). Smith (1986) examined 51 electrophoretic loei in South American and Central American populations of the social spider *Anelosimus eximius* Simon and found seven loci segregating. A mean heterozygosity of 0.060 was found, with only two of seven colonies sampled showing within-colony polymorphism. Roeloffs & Riechert (1988) sampled 44 nests from 17 colonies of *Agelena consociata* Denis from Gabon in Africa. In this social spider, the mean genetic distance between nests belonging to different colonies was significantly higher than that among nests of the same colony, suggesting that nests within an area were part of a single panmictic unit. The mean heterozygosity for 22 loci was 0.018 but each nest was polymorphic at 5.5% of loci examined. On the average, one of the seven loci was found to be polymorphic in each nest. Uetz et al. (1986) studied *Metepeira* species of communal-territorial spiders considered "intermediate" in social versus solitary behavior. They found heterozygosity ranging from 0.09 to 0.21 in three species; variation was lowest in the most communal species.

In this report, we determine the extent of genetic variation in additional genera of solitary spiders from the United States. If solitary spiders can be shown to have low to nonexistent genetic variation, then the Lubin-Crozier hypothesis is weakened and explanations other than inbreeding must be invoked.

METHODS

Spiders were caught individually and taken alive to the USDA, ARS Biological Control of Inseets Research Laboratory (BCIRL) where they were frozen at −80 °C until electrophoresis was

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Table 1.—Allozyme loci, abbreviations and enzyme classification numbers assigned by the International Union of Pure and Applied Chemistry (1973). A dehydrogenase is designated by DH.

Protein	Enzyme Commission Number	No. of loci encoded	Abbreviation
Acid phosphatase	3.1.3.2	2	ACPH
Adenylate kinase	2.7.4.3	1	ADK
Esterase	3.1.1.1	4	EST
Glyceraldehyde-3-phosphate DH	1.2.1.12	1	G-3-PDH
α-glycerophosphate DH	1.1.1.8	2	α-GPDH
Hexokinase	2.7.1.1	1	HK
Hydroxy-β-butyric acid DH	no number	1	H-β-BDH
Isocitrate DH	1.1.1.42	2	IDH
Malate DH	1.1.1.37	2	MDH
Phosphoglucomutase	2.7.5.1	1	PGM
Phosphoglucose isomerase	5.3.1.9	1	PGI
6-phosphogluconate DH	1.1.1.44	1	6-PGDH
Glutamate oxaloacetate transaminase	2.6.1.1	2	GOT

performed. The *Schizocosa* species were taken at Sand Ridge State Forest (*S. ocreata* and *S. stridulans*) or at Chautaqua National Wildlife Refuge (*S. rovneri*) in Mason County, Illinois. *Phidippus clarus* and *M. galathea* were collected from the University of Missouri's Tucker Natural Prairie Preserve, a tall grass prairie remnant located along Interstate Highway 70 in Callaway County, Missouri (Drew 1947).

Starch gel electrophoresis and histochemical staining were performed as described by Steiner & Joslyn (1979), to reveal the proteins listed in Table 1. Protein abbreviations used in this study are also listed there. Sigma starch (Sigma Chemical Co., St. Louis, Missouri) was used at a concentration of 12%, and the 1 cm thick gels were horizontally-sliced into 1 mm thick gel slices for differential enzyme staining.

Two electrode-gel buffer systems were used (Steiner & Joslyn 1979). The first consisted of a LiOH/Boric acid (pH 8.2) electrode buffer and a Trizma base/citric acid (pH 8.4) gel buffer on which *PGI*, *PGM*, *EST* and *ACPH* were stained. The second consisted of a continuous Trizma base/citric acid electrode buffer (pH 8.1) and gel buffer (pH 8.5) system on which were stained *GOT*, *6-PGDH*, *HK*, *ADK*, *α-GPDH*, *IDH*, *G-3-PDH*, and *H-β-BDH*. The allelic bases of the observed isozyme variations were determined using traditional crossing methods to study inheritance patterns and are presented elsewhere (Steiner & Greenstone, 1991). Allele homologies among the different species were not determined.

The derived genetic statistics are as follows. Average number of alleles/locus (A/L) is determined by counting the total revealed by electrophoresis across all loci within a species and dividing by the number of loci examined. The percent of loci polymorphic (%LP) is determined by dividing those which are segregating for two or more alleles by the total number of loci examined within a species. The mean observed percent heterozygosity (%H) is determined by counting the total number of heterozygotes across all loci in a species, dividing by the product of the number of specimans analyzed × number of loci analyzed, and taking the dividend × 100. In general, as the allele frequencies at a locus become equal, the higher the %H will be. High numbers of alleles (high A/L) and of loci polymorphic (high %LP) may be indicative of mutation rate or heterotic selection but do not necessarily correlate with %H. Because of the low numbers of individuals analyzed for each species in this study, we do not presume gene frequencies will be in Castle-Hardy-Weinberg equilibrium, which could also affect expected heterozygosity estimates, since these numbers may be biased due to sampling error.

RESULTS

A total of 21 isozyme loci coding for 13 enzymes was observed across all species. Table 2 indicates each species' genetic profile. The number of loci observed within a species differs from one species to the next, depending on what could be resolved and interpreted. Thus in each of the *Schizocosa* spp. 12 enzyme loci are examined for

Table 2.—Summary of genetic variability for five American Midwest species of arachnids. In the variable loci, a maximum of only 2 alleles were observed for any one locus except where indicated in parentheses. The number of heterozygotes observed are listed over the sample size for that locus. Designations for loci which show no or poor results are indicated for those wishing to pursue genetic investigations of these or other spider species. Abbreviations: B = blurry, not scorable; NA = not analyzed; NP = not present after staining; V = variable but not scorable; see Table 1 for locus abbreviations.

Protein locus	*Schizocosa ocreata*	*Schizocosa rovneri*	*Schizocosa stridulans*	*Phidippus clarus*	*Metaphidippus galathea*
ACPH-1	NA	NA	NA	1/22	0/27
ACPH-2	NA	NA	NA	2/22	0/27
ADK-1	NA	NA	NA	0/22	5/27
EST-1	0/10	2/11	0/5	NP	9/27
EST-2	0/10	2/10	0/5	V, B	V, B
EST-3	NP	NP	NP	V, B	V, B (3)
EST-4	NP	1/10	NP	NP	V, B
α-GPDH-1	0/10	3/12	0/5	0/22	11/27
α-GPDH-2	NP	NP	NP	NP	0/27
GOT-1	B	B	B	0/22	9/27
GOT-2	B	B	B	0/22	0/27
G-3-PDH	0/10	0/12	0/5	0/22	0/27
H-β-BDH	NA	NA	NA	0/22	NA
HK-1	0/10	1/12	0/5	0/22	0/27
IDH-1	0/10	NA	1/5	2/22	4/27
IDH-2	0/10	0/12	0/5	NP	0/27
MDH-1	4/10	4/12	1/5	0/22	0/27
MDH-2	0/10	0/12	1/5	0/22	0/27
PGI	0/10	1/12	0/5	B	0/27
PGM	1/10	0/9	0/4	2/22	0/27
6-PGDH	1/10	4/12 (3)	0/5	0/22	6/27
Alleles/locus	1.25	1.66	1.25	1.20	1.25
% polymorphic	16.60	66.70	25.00	37.50	45.00
% heterozygosity	5.00	12.50	5.00	2.30	9.60

variation compared to 16 in *P. clarus* and 20 in *M. galathea*. The most variable systems were the esterases, but band overlap between loci on the same gel slice sometimes prevented accurate scoring of a particular esterase system. Only loci which could be reliably scored as heterozygous or homozygous were used to develop genetic profiles. Thus *ADK-2* and *HK-2* were not scored and were ignored as they showed up inconsistently and then with only a trace of activity on the gels. Only those esterase loci which could be clearly seen and scored for heterozygosity were included in the final estimate for percent loci polymorphic (Table 2).

At most, we did not observe more than two alleles at any polymorphic locus in any one species with the exception of *EST-3* in *M. galathea* and *6-PGDH* in *S. rovneri* which had 3 alleles each (parentheses, Table 2). The average number of alleles/locus ranged from 1.20–1.66. The percent of loci segregating for two or more alleles ranged from 16.6–66.7 with an average around 38.67%. The mean observed heterozygosity ranged from 5.0–12.5 depending on the species examined, with an overall mean of 6.8.

DISCUSSION

The percent of loci polymorphic which we found in the Missouri *P. clarus* is listed in Table 2 and is 37.5%, very similar to the average of 41.6% observed for all 7 *Phidippus* species in the study by Terranova & Roach (1987). However, our average heterozygosity is only 2.3% for *P. clarus* and does not come close to the average variability of 11.8% seen in the South Carolina populations of this species.

The low levels of variation we observe in the Missouri *P. clarus* is in sharp contrast to that observed in South Carolina. A third as many loci are polymorphic in the 12 loci Terranova and Roach examined compared to the 16 loci we can score for the presence of variation. The variation

observed in the South Carolina population occurs at *PGI*, *AAT*, *IDH-1*, *MDH-1*, *MDH-2* and *amylase*, while that occurring in Missouri is at the *PGM*, *IDH-1*, *EST-2*, *EST-3*, *ACPH-1* and *ACPH-2* loci. The lower heterozygosity seen in Missouri *P. clarus* is one fifth that of South Carolina and is probably due to the polymorphic loci in the Missouri population having lower gene frequencies for alternative alleles.

Such differences in population genetic structure are indicative of adaptive processes at work, and leave room for further question and study. It may be that Midwest populations suffer more often from severe, climatically induced, bottlenecks in population density which are a consequence of harsher winters. Even cultural practices of farmers might play a role if insecticide use is higher in, say, the Midwest, acting to reduce genetic variation through direct selection pressure. These possiblities could explain the differences we observe in Missouri *P. clarus* and can be contrasted with effects due to more inherent phenomena sueh as breeding structure. For example, Lubin (pers. commun.) points out that low levels of variation might be a consequence of breeding system as seen in solitary wasps.

Given the small sample sizes, the five Midwest species of solitary spiders have almost four times the genetic variation observed in *A. wau* by Lubin & Crozier (1985). Other solitary spiders, including the genera *Meta* (Pennington 1979), *Nesticus* (Cesaroni et al. 1981), and *Araneus* (Manchenko 1981), have relatively high levels of genetic variability reflected as isozyme polymorphism as well. These results tend to support the Lubin-Crozier hypothesis. Testing further the robustness of the hypothesis requires more electrophoretic studies of social spiders from more diverse geographic areas and disparate temperate zones and a study of solitary spiders from the area where *A. wau* is endemic. Certainly the range of genetic variation can vary greatly within a genus as we see here in *Schizocosa* and as Terranova & Roach (1987) observed in *Phidippus*. A more meaningful approach might be to look at closely related social and non-social spiders (e. g., see Smith 1987).

Finally, we would point out the significance of having similar levels of variation in *S. stridulans* and *S. ocreata*. These species often occur sympatrically and may share certain life history strategies. Similarities in variability between sympatrically-occurring species is consistent with the idea that micro-evolutionary or adaptive processes transcend species status.

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